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Rheumatoid Arthritis among Women in the Agricultural Health Study: Risk Associated with Farming Activities and Exposures

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PURPOSE: Farming has been associated with increased risk of rheumatoid arthritis (RA) in some studies, but specific causes have not been identified. We studied risk factors for RA in the Agricultural Health Study, a cohort of over 57,000 licensed pesticide applicators and their spouses.
METHODS: We used a nested case-control design, limited to female participants. Physician-confirmed cases ($n = 135$) were matched to five controls each ($n = 675$) by birth date. We used logistic regression, adjusting for birth date and state to examine associations, as estimated by odds ratios (OR) and 95% confidence intervals (CI).
RESULTS: Risk of RA was not associated with mixing or applying pesticides overall or with any pesticide class, nor did it vary by number of days or years of use. Certain pesticides were associated with small non-significantly increased risks, including lindane (OR = 1.8, 95% CI: 0.6–5.0). RA risk was associated with welding (OR = 2.1, 95% CI: 0.8–5.4), albeit imprecisely, but not with solvents or sunlight.
CONCLUSIONS: We did not identify any strong risk factors for RA. Because of the severe disability associated with this relatively common disease, further investigation into causes is warranted both in the Agricultural Health Study and elsewhere.
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KEY WORDS: Rheumatoid Arthritis, Autoimmune Diseases, Autoimmunity, Pesticides, Farming, Occupation.

INTRODUCTION

Rheumatoid arthritis (RA) affects approximately 1% of the United States population, and as many as 2%–3% of those over age 60 (1, 2). Women are more likely to be affected than men, for unknown reasons (3). The course of the disease varies widely, but is generally associated with progressive disability and early mortality (1).

Several epidemiologic studies have reported increased risk of RA among farmers (4–8). Use of pesticides has been associated with slightly increased risk of RA (20%–30% increases) (4, 6); however, specific pesticides have not been studied epidemiologically. The prevalence of antinuclear antibodies (ANAs), a serologic expression of autoimmunity that is not specific to RA, has been associated with residence on a farm among women, as well as with exposure to insecticides including organochlorines, carbamates, and pyrethroids, and exposure to phenoxyacetic acid herbicides (9). Several organophosphate insecticides have been implicated as having toxicologic properties relevant to systemic autoimmunity, including malathion (10) and chlorpyrifos (11).

Case ascertainment and exposure assessment are major difficulties in studying farming exposures as potential causes RA. Self-reporting of RA has been shown to be extremely unreliable in other studies, with confirmation as low as 21%–22% (12, 13). With case ascertainment often requiring extensive validation through medical records, case groups for epidemiologic studies tend to be small. Farming occupation and farm residence are not common in most study populations, and exposures to specific pesticides are even less frequent. The combination of small case groups and infrequent exposures can hinder the informativeness of such studies.

The Agricultural Health Study, a cohort of licensed pesticide applicators and their spouses in Iowa and North Carolina, provides an excellent opportunity to study the health effects of pesticide exposures. Because this population affords a large number of persons involved in farming, the effects of pesticides can be investigated in relation to relatively rare diseases. We investigated pesticide use and other farm-related activities and exposures as possible risk

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Selected Abbreviations and Acronyms

ANA = antinuclear antibody
CI = confidence interval
DDT = dichlorodiphenyltrichloroethane
OR = odds ratio
RA = rheumatoid arthritis

factors for RA among women in the Agricultural Health Study.

METHODS

The Agricultural Health Study

Individuals of any age applying for certification to apply restricted use pesticides in Iowa or North Carolina from 1993 to 1997 were invited to enroll in the Agricultural Health Study (14). Approximately 52,000 private applicators, typically farmers, were enrolled (92% of those eligible); the private applicators were primarily male (2.6% female). A take-home packet contained an invitation for the spouse to enroll and a questionnaire. The number of spouses who agreed to participate in the study and completed the questionnaire was 32,347 (76% of those eligible).

Confirmation of RA Cases in the Agricultural Health Study

We validated RA diagnosis among women who had self-reported RA during a phase II (5 year follow-up) interview for the AHS before December 2003 (24,514 women had responded to this interview). The phase II interview for women (administered to both applicators and spouses) contained several questions about RA symptoms, testing, and age at diagnosis. We targeted the RA validation effort to subgroups of women based on issues bearing on the feasibility of obtaining physician confirmation (e.g., time since diagnosis < 10 years vs. longer), and issues reflecting the likelihood that the women truly had RA (e.g., reported ever having a positive "blood test for RA" [i.e., rheumatoid factor], or reported more than one autoimmune disease). Women who did not report having ever had joint swelling for ≥ 6 weeks (a hallmark symptom of RA and one of the American College of Rheumatology criteria for RA diagnosis) were infrequently selected unless they had another factor indicating probable RA (e.g., positive blood test). The 594 women included in the validation effort, and their inclusion subgroups, are shown in the Appendix. We did not include men in the initial validation because of the lack of information with which to target validation efforts, as men in the Agricultural Health Study were not asked specific questions about RA, such as symptoms and tests.

We recontacted each woman to obtain information about her RA diagnosis and to request signed consent to contact her physicians. For women who self-confirmed their RA diagnosis in the validation interview, we requested information by mail from up to three physicians: (1) the woman's regular physician, who was providing treatment for RA; (2) a rheumatologist (if the regular physician was not a rheumatologist, but there was a rheumatologist that the woman had seen in the past 5 years); and (3) the physician who made the diagnosis of RA (if different from those in 1 and 2). Specific information from the woman's medical history pertaining to RA was elicited from physicians in the form of a checklist, including information on the patient's diagnosis and presence of the American College of Rheumatology classification criteria for RA (15). A woman was classified as having "physician-confirmed" RA if any of her physicians (1) indicated that she had RA by a "yes/no" response, or (2) indicated the presence of at least 4 of 7 American College of Rheumatology classification criteria (15).

Of the 594 women included in the validation process, 136 (23%) cases were physician-confirmed (Table 1). Of the women for whom we received information from any of their physicians ($n = 186$), RA diagnosis was confirmed for 73%. Thirty-three of the physician-confirmed cases (24.3%) were incident between the baseline and phase II

TABLE 1. Validation of female RA cases in the Agricultural Health Study cohort

	Iowa	North Carolina	Total
Final status of total in validation effort*	n (% of 358)†	n (% of 236)†	n (% of 594)†
Did not complete validation interview	32 (9.0)	30 (12.7)	62 (10.4)
Diagnosis not confirmed in validation interview	120 (33.5)	73 (30.9)	193 (32.5)
Diagnosis confirmed in validation interview but consent for physician contact not received	62 (17.3)	67 (28.4)	129 (21.7)
Diagnosis confirmed in validation interview but no response from any physician	13 (3.6)	11 (4.7)	24 (4.0)
Diagnosis confirmed in validation interview but physician did not confirm diagnosis	38 (10.6)	12 (5.1)	50 (8.4)
Physician-confirmed RA‡	93 (26.0)	43 (18.2)	136 (22.9)

*Each group is mutually exclusive.
†Total includes those women who self-reported RA in the baseline or phase II interviews who were contacted for validation; does not include all self-reported RA in Agricultural Health Study.
‡Physician-confirmed by 'yes/no' question about diagnosis or by report of 4 of 7 American College of Rheumatology classification criteria.

interview. For prevalent cases, the duration between the diagnosis and the baseline interview was less than 5 years for 30 cases (22.1%), from 5 to within 10 years for 25 cases (18.4%), and 10 years or longer for 48 cases (35.3%). The ages at the time of diagnosis for physician-confirmed cases ranged from 1 to 73 years (Table 2). Sixty-four percent of the physician-confirmed cases had tested positive for rheumatoid factor (as reported by either the woman or her physician), which is similar to other populations (16, 17).

We considered a more liberal definition of RA than physician-confirmed only, such as inclusion of all 339 women who self-confirmed their RA diagnosis during the validation interview. Of those women, 210 (61.9%) provided consent for us to contact their physicians. Upon further examination of heterogeneity within the self-reported RA cases, we suspected that women who did not provide consent for physician contact included false positive cases, as they were significantly less likely to have been diagnosed by or ever treated by a rheumatologist, were less likely to be currently taking prescription medications for RA, and reported having ever taken fewer different prescription medications for RA than those who consented to physician contact. Based on these discrepancies, we decided to limit our definition of confirmed RA to the "gold standard" of physician-confirmed cases.

Nested Case-Control Study Population

Physician-confirmed RA cases formed the case group for our case-control analysis. Because most of the exposures of interest were relevant to adulthood, we excluded one confirmed case who had been diagnosed at 1 year of age. Cases (n = 135) were matched to five controls each (n = 675) by birth date, within one year. Controls were selected from among women in the Agricultural Health Study who had completed the phase II interview (n = 24,514; the same group from which cases were validated), except those who had reported any systemic autoimmune disease (RA, scleroderma, systemic lupus erythematosus, or Sjögren's syndrome) at either the baseline or phase II interview. The rationale behind this exclusion was based on the fact that we did not attempt to validate every participant who self-reported RA diagnosis, in addition to the difficulty of diagnosis and potential etiologic overlap of systemic autoimmune diseases.

TABLE 2. Characteristics of female RA cases* in the Agricultural Health Study

Characteristic	Cases (n = 136) n (%)
Age at diagnosis (years)	
< 25	8 (5.9)
25–39	39 (28.7)
40–54	53 (39.0)
55–69	33 (24.3)
70 or older	3 (2.2)
Rheumatoid factor	
Positive	87 (64.0)
Negative	49 (36.0)

*Physician-confirmed cases.

Exposure Information

Information on demographics, farming history, pesticide use, and residential, lifestyle, and occupational factors were available from the Agricultural Health Study questionnaires administered at the baseline (questionnaires are available at www.aghealth.org, last accessed September 19, 2005). The questionnaire elicited information about pesticides that were ever personally mixed or applied by the respondent in their lifetime for use on the farm, in the home or garden, or in commercial application (without designating between these different uses). Information of interest included use of 49 specific pesticides (note: permethrin formulations for crop and animal applications were categorized together), and duration and frequency of use for all pesticides combined. Other factors of interest included farming activities and exposures in the longest job held off the farm.

Statistical Analyses

Associations between exposures and the risk of RA were estimated using unconditional logistic regression to generate odds ratios (OR) and 95% confidence intervals (CI), adjusting for birth date (the matching factor) and state (Iowa or North Carolina). After consideration of pack-years of cigarette smoking (0, 0.25–19.5, and $\geq$ 20 pack-years) as a potential confounder in all models, estimates were not importantly affected ($>$ 10% change); thus, final models do not contain the smoking variables.

Several subanalyses were conducted. To evaluate the internal consistency of our results, we conducted analyses stratified by state. Analyses limited to incident cases who were diagnosed within or after the year of enrollment in the Agricultural Health Study (n = 33), and all controls were conducted to check that any observed associations persisted in models in which the exposure was reported prior to disease diagnosis. Similarly, analyses limited to cases who were diagnosed within 5 years before or anytime after their year of enrollment in the Agricultural Health Study (n = 63), and all controls were conducted to check that any observed associations persisted in models for which the reported "ever" exposure was more likely to have occurred prior to disease diagnosis than for more distantly diagnosed cases. Analyses stratified by rheumatoid factor positivity were conducted to evaluate whether any observed associations were limited to one case group.

RESULTS

Of the physician-confirmed RA cases included in the nested case-control study, the median age at diagnosis was 47 years. Only 4 cases and 24 controls were applicators; the remainder were spouses. There were few demographic differences between cases and controls (Table 3), although cases were slightly more likely to reside in North Carolina (OR = 1.3). The risk of RA was associated with pack-years of smoking, with 60% increased risk among women who smoked 20 pack-years or more; however, the risk did not follow a monotonic trend. The pack-years effect was confined to past smokers (for the comparison of 20 or more pack years with never smoked, OR = 2.8, 95% CI: 1.1–7.0 among past smokers and OR = 1.0, 95% CI: 0.4–2.6 among current smokers). Current smokers were at slightly increased risk of RA (OR = 1.3) when pack-years were not taken into account, whereas past smokers were not. RA cases were more likely than were controls to be currently overweight (OR = 1.4, 95% CI: 0.9–2.1), which could be a consequence of disability resulting from the disease. Living on a farm before age 18 was not associated with the risk of RA, nor did the risk vary by duration or age at the time of farm residence.

TABLE 3. Demographic and lifestyle factors and the risk of RA among women in the Agricultural Health Study

Characteristic	Cases (*n* = 135)* *n* (%)†	Controls (*n* = 675) *n* (%)†	OR (95% CI)‡
State of residence			
Iowa	92 (68.2)	494 (73.2)	1.0
North Carolina	43 (31.9)	181 (26.8)	1.3 (0.9–1.9)
Race			
White	131 (97.8)	649 (97.9)	1.0
Non-White	3 (2.2)	14 (2.1)	0.9 (0.3-3.3)
Education			
Did not finish high school	10 (7.5)	21 (3.2)	1.0
High school graduate	52 (38.8)	278 (42.1)	0.8 (0.5–1.3)
Some college	36 (26.9)	178 (26.9)	0.9 (0.5–1.5)
College graduate or more	22 (16.4)	109 (16.5)	0.8 (0.4–1.6)
Cigarette smoking			
Never	94 (72.9)	493 (76.2)	1.0
Past smoker	23 (17.8)	109 (16.9)	1.1 (0.6–1.8)
Current smoker	12 (9.3)	45 (7.0)	1.3 (0.7-2.6)
Pack-years of cigarettes			
0	94 (73.4)	494 (77.2)	1.0
0.25 to 19.5	21 (16.4)	107 (16.7)	1.0 (0.6–1.7)
⩾ 20	13 (10.2)	39 (6.1)	1.6 (0.8-3.2)
Overweight (BMI ⩾25)§	66 (61.7)	257 (54.0)	1.4 (0.9-2.1)
Years lived or worked on a farm			
0 to 10	7 (5.4)	36 (5.6)	1.0
11 to 30	41 (31.5)	190 (29.4)	1.2 (0.5-3.0)
> 30	82 (63.1)	420 (65.0)	1.1 (0.4-2.6)
Lived on farm for	87 (66.4)	410 (63.8)	1.1 (0.8–1.7)
More than 10 years			
Before age 18			

*The nested case-control study included all physician-confirmed cases except one who was diagnosed in infancy.
†Frequencies of characteristics do not always sum to the total number of cases and controls because of missing data.
‡All estimates are adjusted for birth date (matching factor) and state.
§Body mass index (BMI) = (weight in kilograms)/(height in meters^2).

RA risk was not associated with having ever personally mixed or applied pesticides (Table 4), nor did it vary by the number of days or years a woman participated in these activities. Use of herbicides was not associated with RA risk, nor were insecticides overall or the broad insecticide groupings of organophosphates, organochlorines, or carbamates. Elevated ORs were observed for some of the 49 specific pesticides (full set of results not shown), although no estimate was statistically significant, including the insecticides lindane (OR = 1.8), toxaphene (OR = 2.3), malathion (OR = 1.3), DDVP (OR = 1.4), and the herbicide imazethapyr (OR = 1.5). DDT use was not associated with the risk of RA (OR = 1.0). The broad grouping of phenoxyacetic acid herbicides was associated with a significantly decreased risk of RA (OR = 0.5), specifically due to an association with 2,4-D (other herbicides included in this category were 2,4,5-T and MCPA); several other major herbicide classes and specific herbicides showed similar inverse associations with RA, including triazines and thiocarbamates.

Most nonpesticide farming activities and other occupational exposures were not associated with RA (Table 5). 85% of women in the study had worked in jobs off the farm. Elevated odds ratios were observed for both welding on the farm and in off-farm jobs, based on small numbers of exposed subjects, and any welding was associated with 2.1-fold increased risk of RA (6 exposed cases, 16 controls, 95% CI: 0.8–5.4). Elevated, but imprecise odds ratios were also observed for exposure in off-farm jobs to engine exhaust, gasoline, solder, drills, and silica dust. There was virtually no association with sunlight exposure, reported as the number of hours per day spent in the sun during the growing season 10 years prior to the baseline interview.

Some associations differed by state of residence (Table 6). Smoking was associated with increased risk in both states for 20 or more pack-years of cigarettes, although the association was stronger in North Carolina than Iowa. There was some indication that insecticides as a group were associated with increased risk of RA in North Carolina (OR = 1.9) but not Iowa (OR = 1.0). Broad groupings of insecticides associated with elevated ORs in North Carolina included organochlorines and carbamates, although the increases were not statistically significant. Specific pesticides with elevated odds ratios only among North Carolina residents included DDT (OR = 1.8, 95% CI: 0.6-5.2), carbaryl (OR = 2.2, 95% CI: 1.1–4.5), and malathion (OR = 1.6, 95% CI: 0.8–3.5). The modest increased risks observed among the entire study population for welding and lindane were consistent in both states, while data were too sparse to compare

TABLE 4. Agricultural pesticide exposures and the risk of RA among women in the Agricultural Health Study

	Cases (n = 135)*	Controls (n = 675)	
Characteristic	n (%)†	n (%)†	OR (95% CI)‡
Personally mixed or applied pesticides (ever)	88 (65.7)	417 (62.9)	1.2 (0.8–1.7)
Years personally mixed or applied pesticides			
0	46 (40.0)	246 (44.7)	1.0
1 to 10	27 (23.5)	118 (21.4)	1.2 (0.7–2.0)
> 10	34 (29.6)	161 (29.2)	1.1 (0.7–1.7)
Days per year personally mixed or applied pesticides			
0	46 (40.0)	246 (44.4)	1.0
1 to 9	53 (46.1)	222 (40.1)	1.3 (0.8–2.0)
> 9	16 (13.9)	86 (15.5)	1.0 (0.5–1.8)
Type of pesticides used			
Herbicides	54 (40.6)	253 (38.9)	1.1 (0.8–1.6)
Insecticides	66 (50.0)	301 (45.8)	1.2 (0.8–1.7)
Fungicides	4 (3.1)	33 (5.1)	0.5 (0.2–1.6)
Carbamates	52 (39.4)	231 (35.2)	1.2 (0.8–1.7)
Organochlorines	14 (10.9)	63 (10.0)	1.1 (0.6–2.0)
Organophosphates	47 (35.6)	205 (31.3)	1.2 (0.8–1.8)
Phenoxyacetic acids	12 (9.2)	115 (18.0)	0.5 (0.3–0.9)
Thiocarbamates	3 (2.3)	33 (5.1)	0.4 (0.1–1.4)
Triazines	4 (3.1)	44 (6.9)	0.5 (0.2–1.3)
Specific pesticides§			
2,4-D	12 (9.2)	112 (17.6)	0.5 (0.3–0.9)
Alachlor	3 (2.3)	31 (5.0)	0.5 (0.1–1.6)
Atrazine	4 (3.1)	37 (5.8)	0.5 (0.2–1.6)
Carbaryl	50 (38.8)	219 (34.3)	1.2 (0.8–1.8)
Chlordane	3 (2.4)	31 (5.0)	0.5 (0.1–1.6)
Chlorpyrifos	6 (4.7)	35 (5.6)	0.8 (0.4–2.1)
Coumaphos	2 (1.6)	13 (2.1)	0.8 (0.2–3.5)
Cyanazine	2 (1.6)	14 (2.2)	0.8 (0.2–3.4)
DDT	8 (6.5)	38 (6.2)	1.0 (0.4–2.2)
DDVP	5 (3.9)	19 (3.0)	1.4 (0.5–3.9)
Diazinon	12 (9.5)	64 (10.3)	0.9 (0.5–1.7)
Glyphosate	52 (39.1)	222 (34.5)	1.2 (0.8–1.8)
Imazethapyr	5 (3.9)	18 (2.9)	1.5 (0.5–4.1)
Lindane	5 (4.0)	14 (2.3)	1.8 (0.6–5.0)
Malathion	36 (28.4)	150 (23.6)	1.3 (0.8–2.0)
Maneb	3 (2.4)	16 (2.5)	0.8 (0.2–3.0)
Metolachlor	2 (1.6)	26 (4.2)	0.4 (0.1–1.7)
Permethrin	7 (5.3)	35 (5.4)	1.0 (0.4–2.3)
Phorate	2 (1.6)	11 (1.8)	0.9 (0.2–4.3)
Terbufos	3 (2.4)	16 (2.6)	1.0 (0.3–3.4)
Toxaphene	2 (1.6)	4 (0.7)	2.3 (0.4–12.9)

*The nested case-control study included all physician-confirmed cases except one that was diagnosed in infancy.
†Frequencies of characteristics do not always sum to the total number of cases and controls because of missing data.
‡All estimates are adjusted for birth date (matching factor) and state.
§Specific pesticides were presented where the case exposure frequency was >1%; the total number of specific pesticides examined was 49.

risks for DDVP, toxaphene, or imazethapyr. There was some indication of an inverse association with 2,4-D in both states, although the exposure frequency in North Carolina was much lower than in Iowa.

TABLE 5. Nonpesticide occupational activities and exposures and the risk of RA among women in the Agricultural Health Study

	Cases (n = 135)*	Controls (n = 675)	
Characteristic	n (%)†	n (%)†	OR (95% CI)‡
Farming activities§			
Milk cows	3 (2.4)	17 (2.7)	0.9 (0.3–3.2)
Drive trucks	46 (35.9)	215 (33.4)	1.1 (0.8–1.7)
Drive diesel tractors	32 (25.2)	235 (36.4)	0.6 (0.4–1.0)
Drive gasoline tractors	33 (25.8)	178 (27.6)	1.0 (0.6–1.5)
Weld	2 (1.6)	5 (0.8)	2.3 (0.4–11.8)
Grind animal feed	6 (4.7)	31 (4.8)	1.1 (0.4–2.6)
Use gasoline for cleaning	17 (13.3)	114 (17.8)	0.8 (0.4–1.3)
Use other solvents for cleaning	27 (21.1)	150 (23.4)	0.9 (0.6–1.5)
Paint	40 (30.5)	223 (34.7)	0.9 (0.6–1.4)
Contact with animal blood	13 (10.1)	78 (12.1)	0.9 (0.5–1.7)
Hours per day spent in sun during growing season (10 years ago)			
< 1	15 (14.3)	77 (16.2)	1.0
1–2	25 (23.8)	120 (25.3)	1.1 (0.5–2.2)
3–5	39 (37.1)	176 (37.1)	1.1 (0.6–2.2)
6 +	26 (24.8)	102 (21.5)	1.2 (0.6–2.5)
Occupational exposures in job held off the farm#			
Ever had a job off the farm	113 (86.3)	550 (85.0)	1.1 (0.6–1.9)
Asbestos	6 (4.6)	37 (5.7)	0.8 (0.3–2.0)
Cotton dust	2 (1.5)	17 (2.6)	0.5 (0.1–2.1)
Engine exhaust	9 (6.9)	34 (5.3)	1.4 (0.7–3.1)
Gasoline	4 (3.1)	15 (2.3)	1.4 (0.5–4.4)
Grain dust	2 (1.5)	15 (2.3)	0.7 (0.2–3.1)
Lead solder	2 (1.5)	7 (1.1)	1.5 (0.3–7.4)
Mineral or mining dust	2 (1.5)	10 (1.6)	1.0 (0.2–4.8)
Pneumatic drill (vibrations)	2 (1.5)	5 (0.8)	2.1 (0.4–11.1)
Silica/sand dust	3 (2.3)	8 (1.2)	1.9 (0.5–7.4)
Solvents	7 (5.3)	54 (8.4)	0.6 (0.3–1.5)
Welding fumes	4 (3.1)	12 (1.9)	1.8 (0.6–5.6)
Wood dust	2 (1.5)	16 (2.5)	0.6 (0.1–2.7)
X-ray radiation	5 (3.8)	37 (5.7)	0.7 (0.3–1.7)

*The nested case-control study included all physician-confirmed cases except one that was diagnosed in infancy.
†Frequencies of characteristics do not always sum to the total number of cases and controls because of missing data.
‡All estimates are adjusted for birth date (matching factor) and state.
§Conducted at least once per month in summer or winter.
#Self-reported exposure in longest job held off the farm.

In subanalyses restricted to incident cases (n = 33) and all controls (results not shown in tables), elevated ORs were estimated for being overweight (OR = 1.3), 20 or more pack-years of smoking (OR = 1.6), current smoking (OR = 1.8), and welding (OR = 3.1), although all estimates were imprecise due to the very small case group. Elevated odds ratios observed for the entire case group persisted in analyses of incident cases for exposure to the pesticides lindane, toxaphene, DDVP, and imazethapyr. The estimate for malathion was not elevated (OR = 0.7). An inverse association of the risk of RA with 2,4-D use was again observed in the analyses of incident cases (OR = 0.2). Very similar results were observed in the analysis restricted to cases diagnosed within 5 years prior to the baseline interview.

There were no apparent systematic differences in the magnitude of associations with farming exposures between rheumatoid factor positive or negative cases (results not shown). Cases were more likely than controls to be overweight only if they were rheumatoid factor positive (OR = 1.7, 95% CI: 1.0–3.0), perhaps as a consequence of the more severe disease course typical of cases testing positive for rheumatoid factor (18).

DISCUSSION

Several reports have observed increased RA incidence among farmers (4–8), and ours is the first to investigate specific pesticides for their associations with RA. We did not find any strong associations between pesticide exposure and the risk of RA, either for pesticides overall or for broad groupings of pesticide type. These results agree with those of Olsson and colleagues (7), in which RA risk was associated with farming occupation, but not with self-reported pesticide use, and with two other previous studies that found only slightly increased risks (effect estimates of 1.2 and 1.3) associated with pesticide use (4, 6). There was some suggestion in our study that a few specific pesticides may contribute to increased risk of RA; however, we did not have sufficient power to detect these modest increased risks. We had adequate statistical power to detect moderate risks of approximately 2-fold increase (or, conversely, 50% decreased risk) for exposures with at least 10% exposure frequency among controls, as evidenced by our observation of an inverse association with 2,4-D herbicide. Thus, our study was capable of detecting risks of this magnitude for the broad groupings of insecticides and herbicides; the pesticide classes organochlorines; organophosphates; carbamates; and the specific pesticides malathion, carbaryl, diazinon, and glyphosate. The fact that these estimates were not significantly elevated speaks to the probable magnitude of the true associations.

Several major-use herbicides were inversely associated with RA—significantly so for 2,4-D, and similarly for atrazine and alachlor. There have been no previous reports about the potential of these herbicides to contribute to autoimmunity. Suspected immune effects of 2,4-D include immunosuppression,(19, 20) but studies have not focused on the broad spectrum of immune effects including autoimmunity. The similar magnitude of the risk deficits for these herbicides suggests a possible bias resulting from the likely reduction of the women's involvement in farm activities such as pesticide application following disease onset; however, the weakly elevated risk estimates observed for other major-use herbicides such as glyphosate and imazethapyr contradict this theory.

Organochlorine insecticide use was not associated with RA; however, we observed imprecise elevated odds ratios for specific organochlorines, including lindane and toxaphene. We did not observe an association with DDT overall, although there was a moderate, but not-statistically significant, increased risk in North Carolina. It is possible that another risk factor for RA is linked with use of some insecticides in North Carolina (such as silica content of soil in some regions in which specific crops are grown), which could explain why no increased risk was observed in Iowa. North Carolina subjects were on average older than those from Iowa (35% vs. 22% of subjects $\geqslant$ 60 years of age at baseline), and an alternate possibility is that their exposures occurred during an important time window for RA etiology. Chance is also a likely explanation for observed differences between the states.

Evaluation of specific organophosphates indicated weak (not statistically significant) associations with malathion and DDVP. Several experimental studies in animals have indicated effects of malathion on the immune system (21). A study of the insecticide malathion in lupus-prone mice found that oral administration accelerated disease onset and increased the levels of factors indicating autoimmunity including protein in urine, rheumatoid factor, and anti-dsDNA antibody in serum (10); these biologic responses are also relevant to RA. Nevertheless, the inconsistency of the malathion-RA association between states and the non-elevated OR for the association between malathion and incident RA in our study detract from the credibility of the association. Chlorpyrifos was not associated with RA, providing no support for a previous observation of frequent autoantibodies observed among a group of chlorpyrifos-exposed people (11); however, human exposure to chlorpyrifos occurs from multiple sources including diet, and it is possible that our exposure measure did not capture the true variability in exposure.

Farming exposures other than pesticides may be relevant to autoimmune etiology. Other farming exposures including sunlight, dusts (e.g., grain, silica), nonpesticide chemical exposures (e.g., solvents), and viruses have not been extensively researched in relation to autoimmune diseases. We observed an increased risk of RA associated with welding, whether it occurred on or off the farm. This association, although not statistically significant, was observed in both states and in all subanalyses and is in agreement with a previous study that found an 80% increased risk of RA associated with occupation as a mechanic, repairer, sheet metal worker, or welder (7). Several metals that are typical components of welding fumes have been reported to have immune-related effects. Cadmium has been observed to cause antinuclear antibodies in mice upon exposure (22),

TABLE 6. State-specific associations between selected pesticide and other exposures and the risk of RA among women in the Agricultural Health Study (odds ratios [OR] and 95% confidence intervals [CI])

	Iowa			North Carolina		
	Cases (n = 92)	Controls (n = 494)		Cases (n = 43)	Controls (n = 181)	
Characteristic	n (%)*	n (%)*	OR (95% CI)†	n (%)†	n (%)*	OR (95% CI)†
Pack-years of cigarettes						
0	69 (79.3)	379 (79.5)	1.0	25 (61.0)	115 (70.6)	1.0
0.25 to 19.5	13 (14.9)	77 (16.1)	0.9 (0.5–1.7)	8 (19.5)	30 (18.4)	1.2 (0.5–3.0)
⩾ 20	5 (5.8)	21 (4.4)	1.3 (0.5–3.7)	8 (19.5)	18 (11.0)	2.0 (0.8–5.2)
Years lived or worked on a farm						
0–10	5 (5.6)	18 (3.7)	1.0	2 (4.9)	18 (11.4)	1.0
11–30	30 (33.7)	154 (31.6)	0.7 (0.2–2.1)	11 (26.8)	36 (22.8)	2.6 (0.5–13.1)
> 30	54 (60.7)	316 (64.8)	0.7 (0.2–2.0)	28 (68.3)	104 (65.8)	2.1 (0.4–9.8)
Personally mixed or applied pesticides (ever)	60 (65.9)	326 (66.3)	1.0 (0.6–1.6)	28 (65.1)	91 (53.2)	1.8 (0.9–3.5)
Years personally mixed or applied pesticides						
0	31 (39.7)	166 (41.3)	1.0	15 (40.5)	80 (52.6)	1.0
1–10	40 (51.3)	175 (43.5)	1.2 (0.7–2.2)	5 (13.5)	27 (17.8)	1.1 (0.3–3.2)
> 10	7 (9.0)	61 (15.2)	0.9 (0.5–1.6)	14 (37.8)	38 (25.0)	1.8 (0.8–4.0)
Days per year personally mixed or applied pesticides						
0	31 (39.7)	166 (41.6)	1.0	15 (40.5)	80 (52.6)	1.0
1 to 9	22 (28.2)	91 (22.8)	1.2 (0.7–2.1)	13 (35.1)	47 (30.9)	1.6 (0.7–3.8)
> 9	20 (25.6)	123 (30.8)	0.6 (0.3–1.5)	9 (24.3)	25 (16.5)	2.0 (0.8–5.3)
Type of pesticides used						
Insecticides	42 (46.2)	226 (46.4)	1.0 (0.6–1.6)	24 (58.5)	75 (44.1)	1.9 (0.9–3.8)
Herbicides	40 (44.0)	203 (41.9)	1.1 (0.7–1.7)	14 (33.3)	50 (30.1)	1.3 (0.6–2.7)
Fungicides	1 (1.1)	14 (2.9)	0.4 (0.1–2.8)	3 (7.7)	19 (11.4)	0.7 (0.2–2.4)
Organochlorines	6 (6.8)	45 (9.6)	0.7 (0.3–1.8)	8 (20.0)	18 (11.2)	1.9 (0.8–4.8)
Organophosphates	32 (35.2)	151 (31.1)	1.2 (0.8–2.0)	15 (36.6)	54 (32.0)	1.2 (0.6–2.5)
Carbamates	30 (33.0)	165 (33.7)	1.0 (0.6–1.6)	22 (53.7)	66 (39.5)	1.9 (0.9–3.8)
Thiocarbamates	1 (1.1)	20 (4.1)	0.3 (0.04–2.0)	2 (4.8)	13 (7.9)	0.6 (0.1–2.7)
Phenoxyacetic acids	12 (13.3)	102 (21.5)	0.6 (0.3–1.1)	0 (0.0)	13 (7.9)	‡
Triazines	4 (4.4)	40 (8.5)	0.5 (0.2–1.5)	0 (0.0)	4 (2.4)	‡
Specific pesticides						
2,4-D	12 (13.3)	99 (20.9)	0.6 (0.3–1.1)	0 (0.0)	13 (7.9)	‡
Carbaryl	28 (31.8)	160 (33.5)	0.9 (0.6–1.5)	22 (53.7)	59 (36.7)	2.2 (1.1–4.5)
DDT	2 (2.4)	25 (5.4)	0.4 (0.1–1.9)	6 (15.4)	13 (8.5)	1.8 (0.6–5.2)
DDVP	5 (5.6)	18 (3.8)	1.5 (0.6–4.3)	0 (0.0)	1 (0.6)	‡
Glyphosate	38 (41.8)	174 (36.3)	1.3 (0.8–2.0)	14 (33.3)	48 (29.1)	1.3 (0.6–2.8)
Imazethapyr	5 (5.7)	17 (3.7)	1.6 (0.6–4.4)	0 (0.0)	1 (0.7)	‡
Lindane	3 (3.5)	10 (2.2)	1.7 (0.5–6.3)	2 (5.0)	4 (2.6)	2.0 (0.4–11.6)
Malathion	23 (26.4)	114 (23.9)	1.2 (0.7–2.0)	13 (32.5)	36 (22.6)	1.6 (0.8–3.5)
Toxaphene	0 (0.0)	3 (0.7)	‡	2 (5.1)	1 (0.7)	7.2 (0.6–82.2)
Welding (on or off the farm)	5 (5.6)	15 (3.1)	1.9 (0.7–5.2)	1 (2.3)	1 (0.6)	4.0 (0.2–65.8)

*Frequencies of characteristics do not always sum to the total number of cases and controls because of missing data.
†All estimates are adjusted for birth date (matching factor).
‡Effect could not be estimated due to sparse data.

and mercury has been shown to have dual immunosuppressive and immunostimulatory effects depending on the dose, which is a common property among agents suspected to cause autoimmune diseases (21).

A previous report suggested a causal association between substantial solvent use and RA (4), but in our study neither farm nor nonfarm solvent exposures were associated with RA. There is a potential for high exposure to crystalline silica in some agricultural activities (23), and silica exposure may be associated with several autoimmune diseases, including RA (24). While we did not have any assessment of silica on the farm, there was a moderate, albeit imprecise, association with nonfarm silica/sand dust exposure (OR = 1.9). These results for nonpesticide farming exposures are very preliminary, since the detail of information for these exposures was less than for use of pesticides.

Frequency and duration of cigarette smoking contributed to increased risk among past smokers in our study. These results are consistent with previous studies which also found an increased risk of RA associated with smoking (25–33). When pack-years were not taken into account, the risk of RA was higher for current smoking than for past smoking in our study and several others (25, 28, 30), an effect that has been hypothesized to result from smoking causing a short-term decrease in estrogen levels. RA symptoms have been observed to be less frequent in patients during pregnancy when endogenous estrogen levels are high (34), and exogenous estrogen has been shown to suppress collagen-induced arthritis (an animal model for RA) in mice (35, 36). However, there are several other possible pathways by which cigarette smoking might contribute to RA etiology, including oxidative stress (37).

The weak-to-modest associations we observed were with crude exposure metrics such as ever use of a pesticide. More detailed information on frequency and duration of use and intensity of exposure to specific pesticides would be necessary to determine whether stronger associations supporting biologic plausibility are present among the highly exposed. The majority of women in our study were spouses of farmers, as opposed to being farmers themselves. Although most spouses in our study reported using pesticides, it is possible that their exposures are less frequent or at a lower volume than are farmers' exposures; it is also possible that their applications were for home or garden use as opposed to applications on crops or animals. Our results may therefore not be generalizable to farmers and other licensed pesticide applicators. Unfortunately, there were too few female licensed applicators included in our study to examine risk patterns among this group separately; however, exclusion of this group did not change our results. Improved exposure data would also include information on the timing of pesticide use and other exposures relative to RA diagnosis, providing some certainty that exposure occurred before diagnosis. Certainty of the timing of exposure relative to diagnosis is particularly important when examining work-related tasks and exposures because of the possible change in activities resulting from disease-related disability (1).

Although the Agricultural Health Study is a large cohort that includes over 30,000 women, we were able to confirm only a relatively small number of RA cases. This limited the power of our study to identify specific risk factors for RA. Our physician-confirmed case group is likely to include cases that were diagnosed more recently than 10 years ago, because we specifically targeted this subgroup for case validation as cases whose validation would be more feasible. Although not representative of all RA cases, recently diagnosed cases may be more relevant for our analyses, in which we were interested in exposures that occurred prior to diagnosis but for which we did not have information on the timing of exposure. There was some indication that our case group was inclusive of more severe RA cases than would be represented in a general population sample. Among self-confirmed RA cases that consented to physician contact, we were more successful in obtaining physician responses for women with self-reported rheumatoid factor positivity than for those without. It is possible that physicians have greater familiarity with more severe RA cases due to the frequency of patient visits, and were thus more readily able to complete the validation forms. Our results may therefore be more generalizable to a profile of more severe RA than would be encountered in the general population. Nevertheless, our percentage of rheumatoid factor positivity among cases was similar to other study populations of physician-confirmed cases (16).

Despite limitations, our study is the most comprehensive to date in terms of evaluating farming activities and exposures in relation to RA. There is little evidence from this study that pesticides overall or broad pesticide groupings are strongly associated with RA, although our study cannot rule out associations with some specific pesticides. Given the magnitudes of associations with pesticide use observed by us and other researchers, focus should turn toward nonpesticide farming exposures as potential candidates to explain the increased RA incidence observed among farmers. In particular, welding and other metal exposures and silica and other dusts should be studied.

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APPENDIX

Subgroups included in rheumatoid arthritis validation effort*

Group†	Years since diagnosis	Responses in AHS phase II interview	*n* in cohort	*n* in validation effort
1	Any	Reported > 1 autoimmune disease (e.g., RA and either systemic lupus erythematosus, scleroderma, or Sjögren's syndrome)	53	53
2	≤ 10	Swelling‡ and RA blood test§ with positive result	97	97
3	≤ 10	Swelling and RA blood test with negative result	41	41
4	≤ 10	No swelling and RA blood test with positive result	89	89
5	≤ 10	Swelling but no RA blood test done	71	57
6	≤ 10	No swelling and no RA blood test done OR No swelling and RA blood test with negative result	170	50
7	> 10	Swelling and RA blood test with positive result	107	107
8	> 10	EITHER swelling or RA blood test with positive result, not both	103	50
9	Any	Reported RA in phase I but not phase II	558	50

*Among women who responded to the AHS phase II interview (five year follow-up).
†All groups are mutually exclusive.
‡Women responded to the question, "Have you ever had swelling in your wrist, finger, elbow, or knee joints that lasted for six weeks or more?"
§"RA blood test" refers to test for rheumatoid factor.

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